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THE INFLUENCE OF DIETARY PROTEIN AND ENERGY CONCENTRATION ON THE BODY COMPOSITION OF BROILER BREEDER PULLETS

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OPSOMMING: DIE INVLOED VAN VOER-BEPERKING OP LIGGAAMSAMESTELLING VAN GROEIENDE BRAAIKUIKENTEELHENNE

'n Proef, ontwerp om die invloed van verskillende proteïen- en energiepeilbehandelings op vertraging van liggaamsmassatoename van jong braaikuikenteelhenne vas te stel, het karkasontledings op 3 stadia van ontwikkeling gedurende die proef ingesluit. Met geeneen van die behandelings is daarin geslaag om liggaamsmassa te beheer nie. Daar is gevind, wat vetinhoud van die karkasse betref, dat dit oormatig hoog was en dat daar betekenisvolle verskille tussen behandelings was. Vetinhoud van die karkas was afhanklik van die inname van energie. Die vet-inhoud was laer by henne wat lae energie of hoë-proteïenrantsoene ontvang het en wat 'n laer energie-inname tot gevolg gehad het. Daar word voorgestel dat 'n aanvullende ondersoek ingestel moet word om vas te stel in watter liggaamsmassa en liggaamsamestelling die uiteindelike produksievermoë van henne beïnvloed.

SUMMARY

In an experiment designed to test the effect of different protein and energy treatments on body mass restriction of broiler breeder pullets, carcass analyses were carried out on these birds at 3 different stages during the growing period. None of the dietary treatments was successful in limiting body mass at a desirable level. Body fat content was found also to be in excess of a desirable level, although significant differences in body fat and water content were noted. Body fat content was related to the energy intake of the diet, being lower in birds on low energy or high protein diets where the intake of energy was relatively limited. Further research is needed in order to ascertain to what extent body mass and percentage carcass fat should be limited in order to improve reproductivity.

Apart from the fact that body composition and the distribution of chemically or physically defined components in the carcass serve as indices of the nutritive and economic value of meat, the body composition of a bird prior to onset of lay has a bearing on the laying capabilities of that bird. Scott, Nesheim & Young (1969) make it clear that excess fat which is laid down in adipose tissue surrounding the reproductive organs will interfere with optimal egg production, as well as predisposing the birds to greater stress with consequent high mortality during hot weather. Evidence is accruing (Reid, 1971) to show that the body composition of poultry is pliant to nutritional treatments, i.e. body composition yields to dietary manipulation independently of the changes in body mass.

Very little work on the body composition of broiler breeder pullets on a feed restriction programme has been reported. Fuller, Potter & Kirkland (1969), in an attempt to separate the effects of obesity from those of age to sexual maturity on subsequent reproductive performance of White Rock pullets, subjected these birds to a restricted energy intake, decreasing daylength, or both, during the growing period. Controls were full-fed and experienced an increasing daylength. A restriction of energy intake by one-third decreased the carcass fat from 25% on the control diet to 19,9% and 23,8% respectively with increasing and decreasing daylength. Full-fed pullets experiencing the decreasing daylength pattern had carcasses containing 30,1% fat. Because the egg production results paralleled these four restriction treatments, Fuller *et al.* (1969) concluded that the advantages of a decreasing daylength are additive to those of restricted energy.

Donaldson, Combs & Romoser (1956) reported that as the ratio of energy to protein in the ration was widened, the energy intake and carcass fat deposition were increased and the water content of the carcass was decreased. Thus a highly significant positive correlation between calorie/protein ratio of the ration and tissue fat was evident, while highly significant negative correlations were obtained between the calorie/protein ratio and water or protein content of the carcass. The total fat and water content of the carcass remained constant until a calorie (productive energy)/protein ratio of 50 was exceeded. The fat deposited in the carcass was in excess of the water displaced when this ratio in the diets was widened to above 50. Widely different methods of dietary manipulation may be utilized to give different calorie/protein ratios, yet this ratio will still have a high positive correlation with tissue fat in the birds fed these rations (Edwards, Ashour & Nugara, 1971).

As reproductive fitness appears to be related to body composition (Fuller *et al.*, 1969) it would seem necessary to check the body composition of birds on a feed restriction programme. This experiment was designed to ascertain what effect different dietary levels of energy and protein would have on the body composition of broiler breeder replacement pullets. It was hoped that this information might be of value in indicating the most suitable restriction

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Table 1

Composition of the diets fed in the experiment

Component	Diets A	B	C	D	E	F
Protein (%)	16,12	13,95	11,78	16,12	13,95	11,78
Fibre (%)	9,466	9,466	9,467	8,232	8,232	8,232
Energy (kcal/kg)	2260	2260	2260	2978	2978	2978
Calcium (%)	1,530	1,530	1,530	1,530	1,530	1,530
Phosphate (%)	0,701	0,700	0,700	0,700	0,700	0,700
Density (m^3/metric ton)	1,686	1,718	1,742	1,575	1,575	1,607

Table 2

Protein and energy levels used in the experiment, indicating the subdivision of protein treatments at twelve weeks of age

Growing treatment	Period 5 to 12 weeks		Period 13 to 22 weeks	
	Protein (%)	Energy (kcal/kg)	Protein (%)	Energy (kcal/kg)
AA	16,12	2260	16,12	2260
AB	16,12	2260	13,95	2260
AC	16,12	2260	11,78	2260
BB	13,95	2260	13,95	2260
BC	13,95	2260	11,78	2260
CC	11,78	2260	11,78	2260
DD	16,12	2978	16,12	2978
DE	16,12	2978	13,95	2978
DF	16,12	2978	11,78	2978
EE	13,95	2978	13,95	2978
EF	13,95	2978	11,78	2978
FF	11,78	2978	11,78	2978

treatment with regard to the limitation of both body mass and carcass fat percentage.

Procedure

The study was conducted at the University of Natal Research Farm in Pietermaritzburg. A total of 360 White Rock females, 5 weeks of age were placed in 36 pens such that the total mass of the 10 birds in each pen was the same for all pens. The pens were in the form of sunporches with a floor area of one square metre and each pen had one metre each of feeding and watering space. Feed and water were supplied *ad libitum.*

The diets employed in the first 7 weeks of the experiment were at 3 protein levels and 2 energy levels, as shown in Table 1. After 12 weeks of age the treatments were subdivided as shown in Table 2.

The birds were 5 weeks of age in July, 1970, at the initiation of the experiment. As no artificial lighting was available in the pens used, the birds were subjected to the normal seasonal increase in daylight length during the course of the experiment.

Feed consumption was measured each time the body mass of the birds was determined. Body mass of individual birds was measured after a fasting period of 14 hours, and measurements took place at 2 or 3 weekly intervals throughout the growing period. At 12, 17 and 22 weeks of age two birds from each pen (i.e. 6 from each treatment) were removed at random and sacrificed. Carcass analyses for water, fat and protein were carried out on each of these birds.

The carcasses were placed individually in trays in a moisture-extracting oven for 14 days at a temperature of 95°C. The birds were removed after this period, and the percentage water in each carcass was calculated from the wet and moisture-free masses.

The water-free carcasses were passed several times through an electric meat grinder in order to ensure thorough mixing and grinding. Total lipid was determined on a sample of the ground carcass using the method of Du Preez, Wessels, Stokoe & van der Merwe (1971) which entails extraction of fat using chloroform and methanol (2 : 1) followed by diethyl ether. Nitrogen determinations were conducted in triplicate on the fat-free samples using a semi-micro Kjeldahl technique (A.O.A.C., 1966).

All results were converted to percentages on a wet basis. A 6x2 factorial analysis was performed on data at 17 and 22 weeks of age to test for significant differences between treatments. Significance at 12 weeks, due to unequal numbers of observations in each treatment, was obtained from a 6x2 factorial design with two modifications:

The S.S. due to protein

$$= \frac{Y_{i1}^2}{18} + \frac{Y_{i2}^2}{12} + \frac{Y_{i3}^2}{6} - \frac{Y^2}{36}$$

and S.S. due to P X E

$$= \frac{Y_{ij}^2}{n_{ij}} - \text{C.F.} - \text{S.S. (P)} - \text{S.S. (E)}$$

Total and partial correlation coefficients were calculated between carcass components and feed, energy and protein intake. The correlations between carcass components and nutrient intake were determined in an attempt to ascertain the extent to which the carcass components of growing White Rock pullets could be manipulated by dietary means.

Results

Body composition to 22 weeks of age

Mean body composition of birds on each of the treatments are presented in Tables 3, 4 and 5. The tables indicate respectively, body water, body fat on a wet basis, and body protein on a wet basis. Standard errors and coefficients of variation are given for each variate of the 3 age groups under study, and significant differences between treatments are indicated.

At 12 weeks of age neither the low energy diets nor the high energy diets produced significant differences in body water over the 3 protein levels. Significant differences existed, however, between energy levels at each of the 3 dietary protein levels, being lower in birds on the high energy diets.

A number of significant differences in body water between treatments were evident at 17 weeks. These differ-

Table 3

The water content of the carcasses of birds randomly selected from each treatment at three ages during the growing period (Expressed as a percentage of the live body mass)

Age (weeks)	Dietary energy level kcal/kg)	Dietary Protein Level (%)						S.E. C.V.
		16,12			13,95		11,78	
12	2260	65,29^{a}			64,84^{a}		62,48^{ac}	2.208$^{(2)}$
	2978	61,41^{bc}			60.64^{bc}		58,57^{b}	3.52$^{(3)}$
		16,12	13,95	11,78	13,95	11,78	11,78	
17	2260	58.34^{a}	55,18^{ab}	54,52^{ab}	57,23^{ab}	55,77^{abe}	57,08^{ab}	3.322$^{(2)}$
	2978	48,45^{c}	52,62^{bcd}	53,04^{abd}	53,56^{abd}	50,14^{cd}	50,69^{cde}	6.16$^{(3)}$
22	2260	51.84^{ae}	50,68^{ab}	53,12^{ac}	52,37^{ac}	49,99^{ad}	46,56^{ad}	5,468$^{(2)}$
	2978	41,44^{d}	48,66^{ad}	42,60^{bd}	44,82^{bcde}	44,71^{bcde}	52,91^{a}	11,31$^{(3)}$

a, b, c, d, e : Values with the same superscript do not vary significantly ($P < 0{,}05$) within that age group.
(2) : Standard error of a single treatment mean
(3) : Coefficient of variation.

Table 4

The body fat content of the cracasses of birds randomly selected from each treatment at three ages during the growing period (expressed as a percentage of live body mass)

Age	Dietary energy level (kcal/kg)	Dietary protein level (percentage)						S.E. C.V.
		16,12			13,95		11,78	
12	2260	14,25^{a}			15,26^{a}		16,56^{ab}	1,977$^{(2)}$
	2978	17,92^{b}			18,27^{b}		21,33^{c}	11,77$^{(3)}$
		16,12	13,95	11,78	13,95	11,78	11,78	
17	2260	19,84^{a}	22,79^{ac}	23,44^{ac}	21,45^{ac}	22,12^{ac}	21,77^{ac}	2,806$^{(2)}$
	2978	27,85^{b}	25,53^{bc}	24,78^{bc}	23,88^{abc}	27,91^{b}	25,96^{bc}	11,72$^{(3)}$
22	2260	24,73^{ab}	28,09^{abcde}	23,47^{a}	26,15^{abc}	27,67^{abcd}	29,93^{abcdef}	4,617$^{(2)}$
	2978	36,26^{f}	32,31^{bcef}	35,81^{ef}	33,17^{cef}	34,37^{def}	27,04^{abcd}	15,43$^{(3)}$

a......f; (2), (3): See footnotes to Table 3

Table 5

The protein content of the carcasses of birds randomly selected from each treatment at three ages during the growing period (expressed as a percentage of live body mass)

Age (weeks)	Dietary energy level (kcal/kg)	Dietary protein level (percentage)						S.E. C.V.
		16,12			13,95		11,78	
12	2260	18,13^{a}			18,05^{a}		18,24^{a}	1,417$^{(2)}$
	2978	17,50^{a}			17,71^{a}		17,18^{a}	7,95 $^{(3)}$
		16,12	13,95	11,78	13,95	11,78	11,78	
17	2260	18,21^{a}	18,20^{a}	17,98^{a}	17,61^{a}	17,91^{a}	17,81^{a}	1,714$^{(2)}$
	2978	17,92^{a}	17,24^{a}	17,96^{a}	18,34^{a}	17,17^{a}	18,69^{a}	9,56$^{(3)}$
22	2260	17,88^{ab}	18,36^{ab}	17,85^{ab}	17,91^{ab}	17,87^{ab}	16,64^{a}	1,422$^{(2)}$
	2978	18,37^{ab}	17,86^{ab}	17,38^{ab}	19,15^{b}	16,26^{a}	19,08^{b}	8,06$^{(3)}$

a, b; (2), (3) : See footnotes to Table 3.

ences in body water between treatments were evident at 17 weeks. These differences occurred in birds on the high energy diets, and also in birds within protein treatments between the high and low energy diets. No significant differences in body water occurred between any of the birds on the low energy diets. This situation was essentially the same as that at 22 weeks, where significant differences occurred in a few instances but only between protein treatments within the high energy diets, and between energy levels within each protein treatment.

A highly significant negative correlation was obtained between body water and body fat. Consequently the results of the body fat content of birds on the different treatments followed an almost equal but opposite pattern to the results of the body water content of these birds. In the case of body fat content, however, Treatment F differed significantly from Treatments D and E at 12 weeks, but at 17 and 22 weeks there were fewer significant differences within the energy levels. Other significant differences proved to be similar to those between the body water contents of birds on the various treatments.

Tests of significance indicated that, throughout the growing period, body protein remained within very narrow limits for most of the energy and protein treatments under consideration.

Correlations between body constituents and nutrient intake

Total and partial correlation coefficients between body water, body fat and body protein, and between these components and feed, energy and protein intake are presented in Table 6.

Table 6

Total and partial correlation coefficients between carcass constituents and nutrient intake, and between body water, body fat and body protein at three ages during the growing period

Age (weeks)	Variate	Body fat	Body protein	Energy intake	Protein intake	Feed intake	Body mass
12	Body water	−0,8720**	0,1608	−0,5594**	0,4381**	0,0542	0,3966*
		(−0,7518**)	(−0,1181)	(−0,1220)	(0,1470)	(0,0463)	(−0,1171)
	Body fat		−0,2240	0,5543**	−0,4358*	−04358*	0,3297
			(−0,1261)	(0,2213)	(0,0403)	(−0,1285)	(−0,1550)
	Body protein			−0,1827	0,1695	0,0566	−0,1137
				(−0,1384)	(0,0438)	(0,1056)	(0,0371)
17	Body water	−0,9268**	−0,3601*	−0,3412*	0,4441**	0,4664**	−0,2501
		(−0,9268**)	(−0,7200**)	(−0,1532)	(0,0136)	(0,1770)	(0,0069)
	Body fat		0,1061	0,3395	−0,4614**	−0,4798**	0,2559
			(−0,6284**)	(−0,0154)	(−0,0118)	(0,0146)	(−0,0298)
	Body protein			−0,0059	0,0238	0,0258	−0,0652
				(−0,1084)	(0,0487)	(0,1481)	(−0,0429)
22	Body water	−0,9427**	0,0022	−0,3070	0,2254	0,2960	−0,3754*
		(−0,9512**)	(−0,2882)	(0,4465**)	(0,1184)	(−0,4260**)	(−0,3050)
	Body fat		−0,0493	0,4149*	(−0,3381)	−0,4232*	0,3821*
			(−0,3062)	(0,4979)	(0,1116)	(−0,4969)	(−0,2490)
	Body protein			0,0393	0,0705	−0,1029	−0,0582
				(0,3092)	(0,2604)	(−0,3511)*	(−0,2436)

*Indicates significance at P < 0,05. **Indicates significance at P < 0,01

The values are grouped according to the 3 age groups under study. Significant differences between variates are indicated at $P < 0,01$ and $P < 0,05$.

At 12 weeks of age, a highly significant negative correlation ($P < 0,01$) was found to exist between body water and body fat on a wet basis. Consequently the values for each of these variates were found to be almost equal but opposite when between-treatments means were analysed. At the same age, energy intake, protein intake and body mass were correlated with body water. Body fat was correlated positively with energy intake and negatively with protein intake.

At 17 weeks of age, both body fat and body protein were significantly correlated with body water. Protein and feed intake were significantly positively correlated with body water, and negatively with body fat.

The significant negative correlation between body water and body fat was evident at 22 weeks of age, as was the negative correlation between feed intake and body fat. Body fat was found to be positively correlated with both energy intake and body mass, this correlation proving to be significant. The negative correlation between body water and body mass which was significant at 12 weeks, was again significant at 22 weeks of age.

Discussion

Body water, body fat and body protein

The fact that the body composition of the birds in this trial proved to be pliant to nutritional changes agrees very favourably with most of the published work on body composition studies with poultry. Amongst others, Hill & Dansky, (1951); Donaldson *et al.*, (1955); Du Preez, Du Plessis & Erasmus (1967) and Edward *et al.* (1971) have all noted the effect of dietary energy and protein levels on body composition of broilers. These effects, as proved in this trial, demonstrate that the percentage of body fat increases as the energy level of the diet increases, or as the protein level of the diet decreases. This is particularly noticeable at 12 weeks, and to a lesser extent at 17 and 22 weeks of age (see Table 4). Except for a few treatment means, the trend of increasing body fat, or decreasing body water, follows the expected values for all the treatments.

The actual values obtained for the percentage of body fat on a wet basis are slightly higher at 22 weeks than the values obtained by Fuller *et al.*, (1969). However, the body masses were considerably higher than those recorded by Fuller. It is conceivable that the major portion of the additional body mass of the birds in the present trial would be body fat.

The protein content of the carcasses remained surprisingly constant between treatments and at different ages. This agrees with the results of Fuller *et al.* (1969) and Edwards *et al.* (1971). Bailey & Zobrisky (1968) state that the animal body is able to maintain to a high degree its essential protein composition, regardless of differences in age, mass or nutrient intake. The present results certainly support this statement.

Reid (1959) noted that a lower plane of nutrition and the resultant retardation of early growth are associated with the prolongation of the life span in a wide variety of animal and insect species. These observations, together with the remarks of Scott *et al.* (1969) that overfat birds do not reproduce efficiently indicate that birds on the high protein, low energy diet in this experiment would be expected to show greatest reproductive fitness in the laying stage. The birds on this treatment (AA) exhibited a significantly lower body fat content than most of the birds on the high energy diets. The body fat content of these birds was also lower than that of all other birds fed low energy diets except for one diet (AC), although these differences were not significant. To keep the body fat content of broiler breeder pullets as low as is possible with an *ad lib.* diet, it would therefore appear that a high protein, low energy diet, i.e. a diet with a low calorie protein ratio, would be required to be fed.

It is difficult to explain the fact that at 22 weeks the birds on Treatment AC showed a body fat content lower than that of the birds on the high protein low energy diet. This was not to be expected on the evidence that a lowering of the protein level in the diet results in an increased fat content (Petersen, Grau & Peak, 1954). However this difference did not prove to be statistically significant.

Correlations between carcass constituents and energy, protein and feed intakes and body mass

The highly significant negative correlation between body water and body fat (percentages, on a wet basis) has been shown to exist in many animals and birds (Brody, 1945; Combs & Robel, 1962; Du Preez *et al.*, 1969). A correlation of $-0,97$ in this trial was obtained by grouping together data from all birds analysed, without regard to age or dietary treatment imposed. It would appear therefore that a very accurate estimation of carcass fat content can be made of carcasses whose water content is known. As the water content of a bird is relatively easy to measure, this relationship would appear to be very useful in carcass composition studies. However, in young rats and chickens where a significant positive correlation between body water and body nitrogen has been demonstrated (Bender & Miller, 1953; De Muelenaere, Quicke & Wessels, 1960; and Combs & Robel, 1962) the negative relationship between body fat and body water might not be as marked as indicated in the present study. It is expected that as fat deposition begins to take place in increasing amounts, with a corresponding "flattening" of the body protein curve, the positive relationship between body water and body nitrogen will diminish and then approximate zero. The low correlation between body water and body protein in the present experiment confirms this fact.

It would therefore appear that the method of Bender & Miller (1953) and De Muelenaere *et al.*, (1960) of estimating carcass nitrogen from body water is suitable only for young rats and chickens. Similarly, estimating percentage fat from percentage water is valid only within limits and these limits must be established separately for ages and species.

A number of the correlations in Table 6 are difficult to interpret. The significant negative total correlation found to exist between body water and body protein at 17 weeks of age was totally unexpected. It is possible that the change of diet at 12 weeks had the effect of temporatily upsetting the normal balance between the various body components as shown by the total correlation coefficient between feed intake and body water, which was significant only at 17 weeks, and by the positive total correlation coefficient (non-significant) between body protein and body fat at the same age.

The percentage fat in the carcasses tended to be positively related to both body mass and energy intake, but negatively related to both protein intake and feed intake at all three ages studied. In support of the positive body mass to body fat correlations, Hill & Dansky (1951), Du Preez *et al.* (1966) and Wethli (1968) found that by increasing the level of dietary energy they obtained increased carcass fat, and this was accompanied by greater body masses. This would also account for the positive relationship between carcass fat and energy intake, due to the fact that these broilers tended to overconsume energy above their energy requirements. According to Dickerson (1947) and Reid (1971), a strong negative relationship between feed conversion and body fat is indicative of low activity and large appetite, which agrees well with the state of the birds in this trial.

The negative correlation between feed intake and carcass fat is an indication of the widely different energy levels used in this experiment. Birds on the high energy treatments consumed less feed than those on the low energy treatments, but nevertheless consumed more energy. Consequently these birds exhibited a higher body fat content than the birds on the low energy diets. Similarly, birds on the low protein treatments consumed less protein but more energy than the birds receiving high protein diets, consequently showing a higher body fat content. This trend was noticeable especially at 12 weeks, but persisted to a lesser degree throughout the growing period.

It is therefore reasonable to assume that the fat content of a carcass is positively related to the amount of energy consumed by the bird, and any method whereby this energy intake can successfully be reduced, will result in a leaner carcass. The birds with a leaner body composition will in turn exhibit more efficient reproductivity (Fuller *et al.*, 1969).

A leaner carcass is, however, not necessarily related to a lower body mass. Cohn (1963) and Reid (1971) observed in rats that the frequency of feeding had a marked effect on body composition, but no effect on body weight gain. Rats ingesting their daily allowance in two meals utilized a far higher proportion of their dietary energy for fattening than did those allowed to nibble continuously. These workers reported that the rats fed *ad lib.* had a carcass fat content 6,4% lower than the others. There are two questions that arise from these observations. Firstly, is body mass the most important variate that should be limited in a broiler feed restriction programme, or should carcass fat content be the criterion governing the method of restriction? The other question concerns the effect on body composition of a skip-a-day, or limited-time feeding schedule as used by many large broiler breeding enterprises. With feeding time being limited it might be expected that the restricted birds will tend to deposit more body fat as a consequence of "gorging" themselves on every occasion that feed is presented.

The present data illustrate the point that a reappraisal of the variate to be limited at point-of-lay is necessary. Birds on Treatments AA and EF at 22 weeks exhibited very similar mean body masses. Carcass fat content differed however by 9,64% (P nearly < 0,01). It would appear obvious from the observations of the effects of obesity on reproductive fitness quoted previously that birds from Treatment AA would be more efficient egg producers than birds from Treatment EF, even though their body masses were so similar at 22 weeks. A reappraisal of the methods and the aims of feed restriction programmes as applied to broiler breeder pullets is clearly required.

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